

What is claimed is:

- Sub. B1
1. An isolated HIV-1 Group O *env* polypeptide having an amino acid sequence consisting essentially of the sequence of Figure 1 (SEQ ID NO:61).
  2. An isolated HIV-1 Group O *env* polypeptide comprising an immunoreactive portion of a polypeptide according to Claim 1.
  3. A polynucleotide encoding a polypeptide according to any of Claims 1 and 2.
  4. An antigen construct comprising a first HIV-1 Group O *env* polypeptide fused to a second HIV-1 Group O *env* polypeptide.
  5. An antigen construct according to Claim 4 wherein the first polypeptide is a gp120 polypeptide, and the second polypeptide is a gp41 polypeptide.
  6. An antigen construct according to Claim 5 wherein a portion of the hydrophobic region of the gp41 polypeptide has been deleted.
  7. An antigen construct according to any of Claims 4, 5 and 6 wherein at least one of the first and second HIV-1 Group O *env* polypeptides is derived from HIV-1 Group O isolate HAM112.
  8. An antigen construct according to Claim 4 wherein the first polypeptide comprises an immunoreactive portion of the gp120 protein of HIV-1 Group O isolate HAM112.
  9. An antigen construct according to Claim 8 wherein the first polypeptide has an amino acid sequence consisting essentially of residues 1 through 520 of the sequence of Figure 1 (SEQ ID NO:61) or a portion thereof.
  10. An antigen construct according to Claim 9 wherein the first polypeptide has an amino acid sequence consisting essentially of residues 476 through 520 of the sequence of Figure 1 (SEQ ID NO:61).

11. An antigen construct according to any of Claims 4, 8, 9 and 10 wherein the second polypeptide comprises an immunoreactive portion of the gp41 protein of HIV-1 Group O isolate HAM112.

12. An antigen construct according to Claim 11 wherein the second polypeptide has an amino acid sequence consisting essentially of residues 521 through 873 of the sequence of Figure 1 (SEQ ID NO:61) or a portion thereof.

13. An antigen construct according to Claim 11 wherein a portion of the hydrophobic region of the gp41 protein of HIV-1 Group O isolate HAM112 is absent from the second polypeptide.

14. An antigen construct according to Claim 11 wherein the deleted portion is that part of gp41 having an amino acid sequence consisting essentially of residues 690 through 715 of the sequence of Figure 1 (SEQ ID NO:61).

15. An antigen construct according to Claim 11 wherein the second polypeptide has an amino acid sequence consisting essentially of residues 47 through 373 of Figure 9 (SEQ ID NO:52).

16. An antigen construct according to Claim 11 wherein the second polypeptide has an amino acid sequence consisting essentially of residues 47 through 245 of Figure 7 (SEQ ID NO:48).

17. An antigen construct according to Claim 11 wherein the second polypeptide has an amino acid sequence consisting essentially of residues 47 through 215 of Figure 5 (SEQ ID NO:58).

18. An antigen construct according to Claim 4 selected from the group consisting of pGO-8PL, pGO-8CKS, pGO-9PL, pGO-9CKS, pGO-11PL and pGO-11CKS, and derivatives, variants and analogs thereof.

19. An antigen construct comprising a fusion of at least one HIV-1 Group O *env* polypeptide with at least one HIV-1 Group M *env* polypeptide.

20. An antigen construct according to Claim 19 comprising a fusion of:
- (a) a first HIV-1 Group O *env* polypeptide;
  - (b) a second HIV-1 Group O *env* polypeptide;
  - (c) a first HIV-1 Group M *env* polypeptide; and
  - (d) a second HIV-1 Group M *env* polypeptide.
21. An antigen construct according to Claim 20 wherein the HIV-1 Group M polypeptides are derived from an HIV-1 isolate of Subtype B.
22. An antigen construct according to Claim 21 wherein at least one of the HIV-1 Group M polypeptides is derived from HIV-1 Group M isolate HXB2R.
23. An antigen construct according to any of Claims 20, 21 and 22 wherein at least one of the HIV-1 Group O sequences is derived from HIV-1 Group O isolate HAM112.
24. An antigen construct according to Claim 20 wherein the first Group O *env* polypeptide and the first Group M *env* polypeptide are both gp120 polypeptides, and the second Group O *env* polypeptide and the second Group M *env* polypeptide are both gp41 polypeptides.
25. An antigen construct according to Claim 24 wherein a portion of the hydrophobic region of at least one of the gp41 polypeptides has been deleted.
26. An antigen construct according to Claim 20 wherein:
- (a) the first HIV-1 Group O *env* polypeptide comprises an immunoreactive portion of the gp120 protein of HIV-1 Group O isolate HAM112;
  - (b) the second HIV-1 Group O *env* polypeptide comprises an immunoreactive portion of the gp41 protein of HIV-1 Group O isolate HAM112;
  - (c) the first HIV-1 Group M *env* polypeptide comprises an immunoreactive portion of the gp120 protein of a first HIV-1 Group M isolate of Subtype B; and
  - (d) the second HIV-1 Group M *env* polypeptide comprises an immunoreactive portion of the gp41 protein of a second HIV-1 Group M isolate of Subtype B.
27. An antigen construct according to Claim 26 wherein the first and second HIV-1 Group M isolates of Subtype B are the same and are HIV-1 Group M isolate HXB2R.

28. An antigen construct according to Claim 27 wherein a portion of the hydrophobic region of the gp41 protein of HIV-1 Group M isolate HXB2R is absent from the second HIV-1 Group M *env* polypeptide.

29. An antigen construct according to Claim 26 wherein:

(a) the first HIV-1 Group M *env* polypeptide has an amino acid sequence consisting essentially of residues 251 through 292 of the sequence of Figure 12 (SEQ ID NO:108); and

(b) the second HIV-1 Group M *env* polypeptide has an amino acid sequence consisting essentially of residues 293 through 599 of the sequence of Figure 12 (SEQ ID NO:108) or a portion thereof.

30. An antigen construct according to Claim 29 wherein the second HIV-1 Group M *env* polypeptide has an amino acid sequence consisting essentially of residues 293 through 492 of the sequence of Figure 12 (SEQ ID NO:108).

31. An antigen construct according to any of Claims 20, 24, 25, 26, 27, 28, 29 and 30 wherein the first HIV-1 Group O *env* polypeptide has an amino acid sequence consisting essentially of residues 1 through 520 of the sequence of Figure 1 (SEQ ID NO:61) or a portion thereof.

32. An antigen construct according to Claim 31 wherein the second HIV-1 Group O *env* polypeptide has an amino acid sequence consisting essentially of residues 521 through 873 of the sequence of Figure 1 (SEQ ID NO:61) or a portion thereof.

33. An antigen construct according to Claim 31 wherein the first HIV-1 Group O *env* polypeptide has an amino acid sequence consisting essentially of residues 476 through 520 of the sequence of Figure 1 (SEQ ID NO:61).

34. An antigen construct according to Claim 33 wherein a portion of the hydrophobic region of the gp41 protein of HIV-1 Group O isolate HAM112 is absent from the second HIV-1 Group O *env* polypeptide.

35. An antigen construct according to Claim 33 wherein the second HIV-1 Group O *env* polypeptide has an amino acid sequence consisting essentially of residues 47 through 373 of Figure 9 (SEQ ID NO:52).

36. An antigen construct according to Claim 33 wherein the second HIV-1 Group O *env* polypeptide has an amino acid sequence consisting essentially of residues 47 through 245 of Figure 7 (SEQ ID NO:48).

37. An antigen construct according to Claim 33 wherein the second HIV-1 Group O *env* polypeptide has an amino acid sequence consisting essentially of residues 47 through 215 of Figure 5 (SEQ ID NO:58).

38. An antigen construct according to Claim 19 selected from the group consisting of pGO-12CKS, pGO-13CKS and pGO-14PL, and derivatives, variants and analogs thereof.

39. An antigen construct comprising a fusion of a first HIV-1 *env* polypeptide, a second HIV-1 *env* polypeptide, and at least one additional HIV-1 polypeptide.

40. An antigen construct according to Claim 39 wherein the first HIV-1 *env* polypeptide and the second HIV-1 *env* polypeptide are each HIV-1 Group O polypeptides.

41. An antigen construct according to Claim 40 wherein the first HIV-1 Group O *env* polypeptide is a gp120 polypeptide, and the second HIV-1 Group O *env* polypeptide is a gp41 polypeptide.

42. An antigen construct according to Claim 41 wherein the first HIV-1 Group O *env* polypeptide comprises an immunoreactive portion of the gp120 protein of HIV-1 Group O isolate HAM112, and the second HIV-1 Group O *env* polypeptide comprises an immunoreactive portion of the gp41 protein of HIV-1 Group O isolate HAM112.

43. An antigen construct according to Claim 42 wherein the first HIV-1 Group O *env* polypeptide has an amino acid sequence consisting essentially of residues 1 through 520 of the sequence of Figure 1 (SEQ ID NO:61) or a portion thereof.

44. An antigen construct according to Claim 43 wherein the second HIV-1 Group O *env* polypeptide has an amino acid sequence consisting essentially of residues 521 through 873 of the sequence of Figure 1 (SEQ ID NO:61) or a portion thereof.

45. An antigen construct according to Claim 43 wherein the first HIV-1 Group O *env* polypeptide has an amino acid sequence consisting essentially of residues 476 through 520 of the sequence of Figure 1 (SEQ ID NO:61).

46. An antigen construct according to Claim 45 wherein a portion of the hydrophobic region of the gp41 protein of HIV-1 Group O isolate HAM112 is absent from the second HIV-1 Group O *env* polypeptide.

47. An antigen construct according to Claim 45 wherein the second HIV-1 Group O *env* polypeptide has an amino acid sequence consisting essentially of residues 47 through 373 of Figure 9 (SEQ ID NO:52).

48. An antigen construct according to Claim 45 wherein the second HIV-1 Group O *env* polypeptide has an amino acid sequence consisting essentially of residues 47 through 245 of Figure 7 (SEQ ID NO:48).

49. An antigen construct according to Claim 45 wherein the second HIV-1 Group O *env* polypeptide has an amino acid sequence consisting essentially of residues 47 through 215 of Figure 5 (SEQ ID NO:58).

50. An antigen construct according to any of Claims 40, 41, 42, 43, 44, 45, 46, 47, 48 and 49 wherein the additional HIV-1 polypeptide is a Group O *env* polypeptide.

51. An antigen construct according to Claim 50 wherein the additional HIV-1 Group O polypeptide comprises an immunoreactive portion of the gp41 protein of HIV-1 Group O isolate HAM112.

52. An antigen construct according to Claim 51 wherein the additional HIV-1 Group O polypeptide has an amino acid sequence consisting essentially of residues 521 through 873 of the sequence of Figure 1 (SEQ ID NO:61) or a portion thereof.

53. An antigen construct according to Claim 52 wherein a portion of the hydrophobic region of the gp41 protein of HIV-1 Group O isolate HAM112 is absent from the additional HIV-1 Group O polypeptide.

54. An antigen construct according to Claim 50 wherein the additional HIV-1 Group O *env* polypeptide has an amino acid sequence consisting essentially of residues 47 through 373 of Figure 9 (SEQ ID NO:52).

55. An antigen construct according to Claim 50 wherein the additional HIV-1 Group O *env* polypeptide has an amino acid sequence consisting essentially of residues 47 through 245 of Figure 7 (SEQ ID NO:48).

56. An antigen construct according to Claim 50 wherein the additional HIV-1 Group O *env* polypeptide has an amino acid sequence consisting essentially of residues 47 through 215 of Figure 5 (SEQ ID NO:58).

57. An antigen construct according to Claim 50 wherein the additional HIV-1 Group O *env* polypeptide has an amino acid sequence consisting essentially of residues 250 through 281 of Figure 17 (SEQ ID NO:120).

58. An antigen construct according to Claim 40 selected from the group consisting of pGO-15CKS and pGO-15PL, and derivatives, variants and analogs thereof.

59. An antigen construct comprising a first HIV-2 *env* polypeptide fused to a second HIV-2 *env* polypeptide.

60. An antigen construct according to Claim 59 wherein the first HIV-2 *env* polypeptide is a gp120 polypeptide, and the second HIV-2 *env* polypeptide is a gp36 polypeptide.

61. An antigen construct according to Claim 60 wherein:  
(a) the first HIV-2 *env* polypeptide has an amino acid sequence consisting essentially of residues 248 through 307 of the sequence of Figure 11 (SEQ ID NO:55) or a portion thereof; and  
(b) the second HIV-2 *env* polypeptide has an amino acid sequence consisting essentially of residues 308 through 466 of the sequence of Figure 11 (SEQ ID NO:55) or a portion thereof.

62. An antigen construct according to Claim 59 which is pHIV-210, and derivatives, variants and analogs thereof.

63. A polynucleotide encoding an antigen construct according to any of Claims 4, 18, 19, 38, 39, 58, 59 and 62.

64. A polynucleotide according to Claim 63 operably linked to a control sequence capable of directing expression in a suitable host.

65. A polynucleotide according to Claim 63 wherein the coding sequence is modified to provide a codon bias appropriate to the expression host.

66. An expression vector comprising a polynucleotide according to Claim 63.
67. A host cell transformed by an expression vector according to Claim 66.
68. A host cell according to Claim 67 wherein the host is *Escherichia coli*.
69. A method for detecting antibodies to HIV-1 in a test sample comprising the steps of:
- (a) combining at least one antigen construct according to any of Claims 4, 18, 19, 38, 39, 58, 59 and 62 with the test sample to form a mixture;
  - (b) incubating the mixture under conditions suitable for formation of complexes between the antigen and antibodies, if any, which are present in the sample and are immunologically reactive with the antigen; and
  - (c) detecting the presence of any complexes formed.
70. A method according to Claim 69 wherein detecting the presence of complexes in step (c) is carried out using an additional antigen construct according to any of Claims 4, 18, 19, 38, 39, 58, 59 and 62 to which is attached a signal-generating compound.
71. A method according to Claim 69 wherein detecting the presence of complexes in step (c) is carried out using an additional antigen construct according to any of Claims 4, 18, 19, 38, 39, 58, 59 and 62 to which is attached a first member of a specific binding pair, and further using an indicator reagent comprising a second member of the specific binding pair to which is attached a signal-generating compound.
72. A method according to Claim 69 wherein detecting the presence of complexes in step (c) is carried out using an antibody directed to the complexes formed in step (b) to which is attached a signal-generating compound.
73. A method according to Claim 69 wherein detecting the presence of complexes in step (c) is carried out using an antibody directed to the complexes formed in step (b) to which is attached a first member of a specific binding pair, and further using an indicator reagent comprising a second member of the specific binding pair to which is attached a signal-generating compound.
74. An immunoassay kit for the detection of antibodies to HIV-1 comprising an antigen construct according to any of Claims 4, 18, 19, 38, 39, 58, 59 and 62.



75. An immunoassay kit according to Claim 74 wherein the antigen construct is a capture reagent.

76. An immunoassay kit according to Claim 74 wherein the antigen construct is an indicator reagent.

77. An immunoassay kit according to Claim 74 wherein the antigen construct is attached to a first member of a specific binding pair, the kit additionally comprising an indicator reagent comprising a second member of the specific binding pair attached to a signal-generating compound.

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